Atp13a2 Cas9-CKO Strategy

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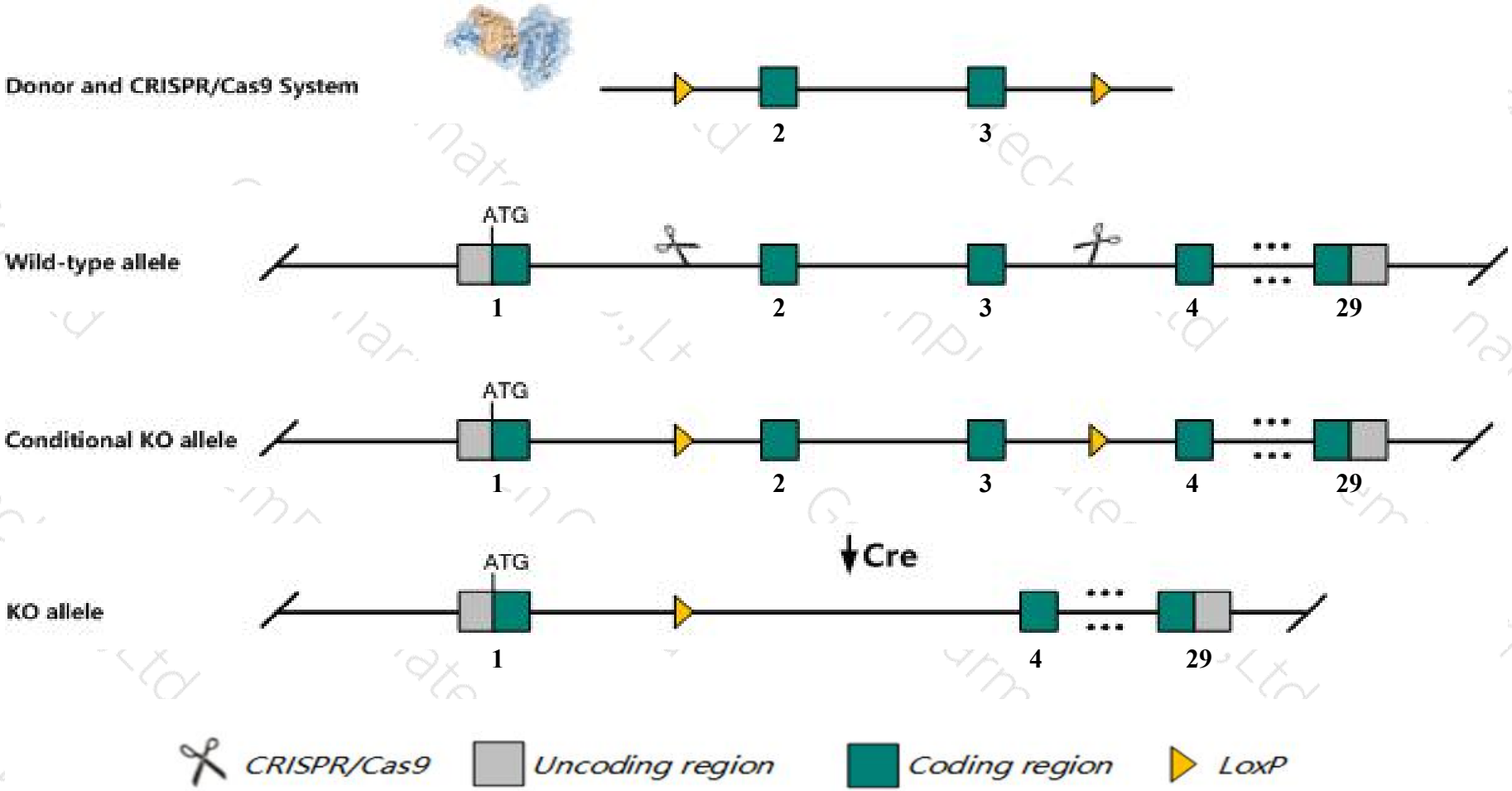
Project Overview

Project Name	***Atp13a2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Atp13a2* gene. The schematic diagram is as follows:



Technical routes

- The *Atp13a2* gene has 9 transcripts. According to the structure of *Atp13a2* gene, exon2-exon3 of *Atp13a2-201* (ENSMUST00000037055.13) transcript is recommended as the knockout region.The region contains 278bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Atp13a2* gene. The brief process is as follows:CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice

➢ According to the existing MGI data,Mice homozygous for a knock-out allele exhibit neuronal ceroid lipofuscinosis, synuclein accumulation and age-dependent sensorimotor deficits.

➢ Transcript *Atp13a2*-204,205,207,209 may not be affected.

➢ The *Atp13a2* gene is located on the Chr4. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.

➢ This Strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Atp13a2 ATPase type 13A2 [Mus musculus (house mouse)]

Gene ID: 74772, updated on 2-Apr-2019

Summary

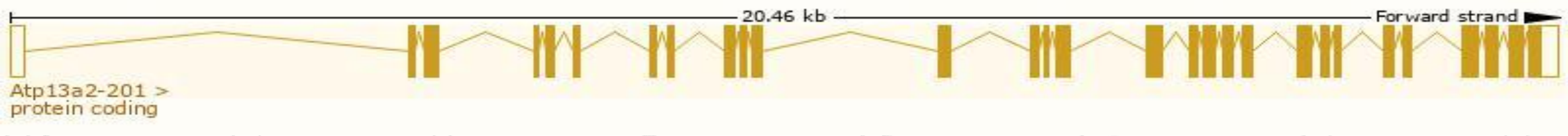
Official Symbol	Atp13a2 provided by MGI
Official Full Name	ATPase type 13A2 provided by MGI
Primary source	MGI:MGI:1922022
See related	Ensembl:ENSMUSG00000036622
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110012E06Rik, AA589443
Expression	Ubiquitous expression in spleen adult (RPKM 76.5), ovary adult (RPKM 68.9) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

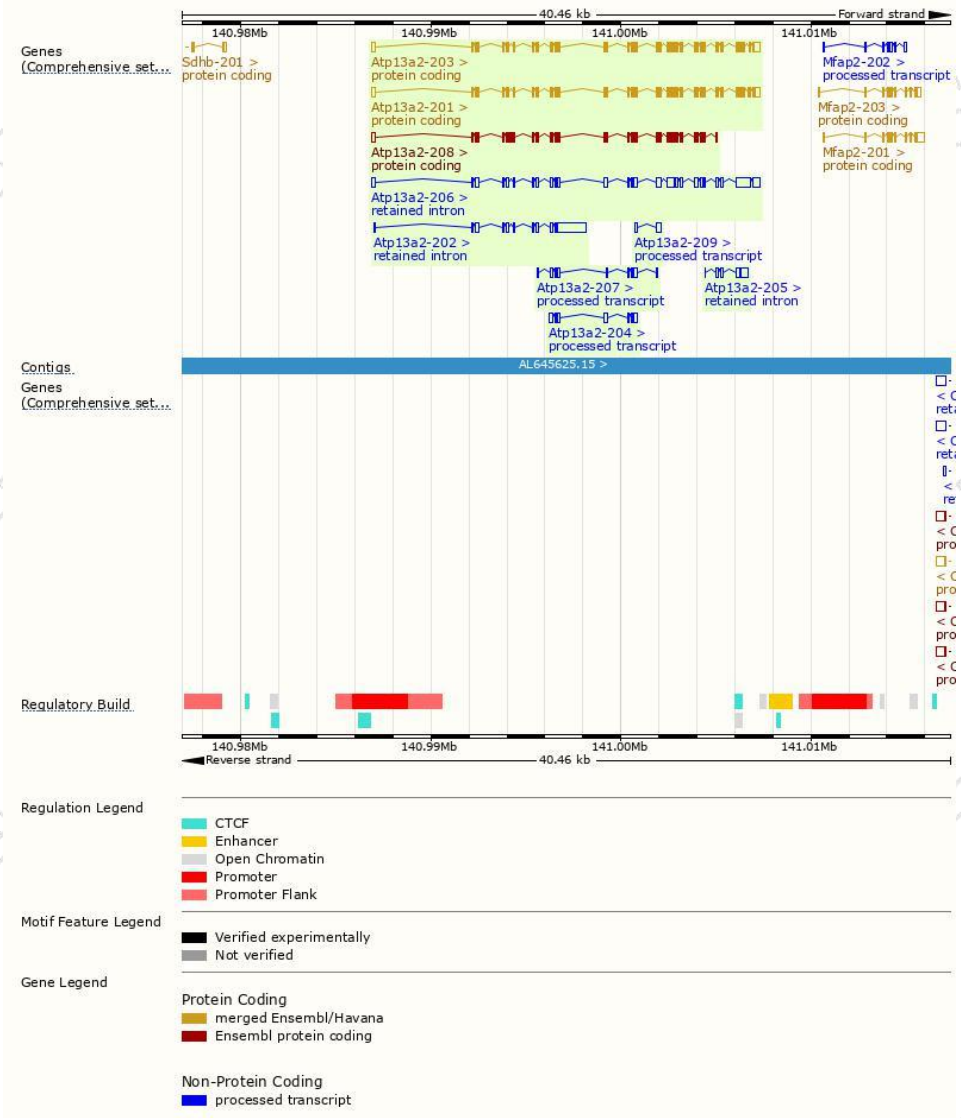
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atp13a2-201	ENSMUST00000037055.13	3956	1169aa	Protein coding	CCDS18859	Q9CTG6	TSL:1 GENCODE basic APPRIS P3
Atp13a2-203	ENSMUST00000127833.2	3882	1115aa	Protein coding	CCDS51345	E9Q2A4	TSL:1 GENCODE basic APPRIS ALT2
Atp13a2-208	ENSMUST00000168047.7	3105	970aa	Protein coding	-	E9PYX5	CDS 3' incomplete TSL:1
Atp13a2-204	ENSMUST00000135117.1	927	No protein	Processed transcript	-	-	TSL:2
Atp13a2-207	ENSMUST00000156995.7	807	No protein	Processed transcript	-	-	TSL:5
Atp13a2-209	ENSMUST00000170797.1	366	No protein	Processed transcript	-	-	TSL:3
Atp13a2-206	ENSMUST00000151517.7	4266	No protein	Retained intron	-	-	TSL:2
Atp13a2-202	ENSMUST00000125797.7	2469	No protein	Retained intron	-	-	TSL:1
Atp13a2-205	ENSMUST00000137630.2	877	No protein	Retained intron	-	-	TSL:3

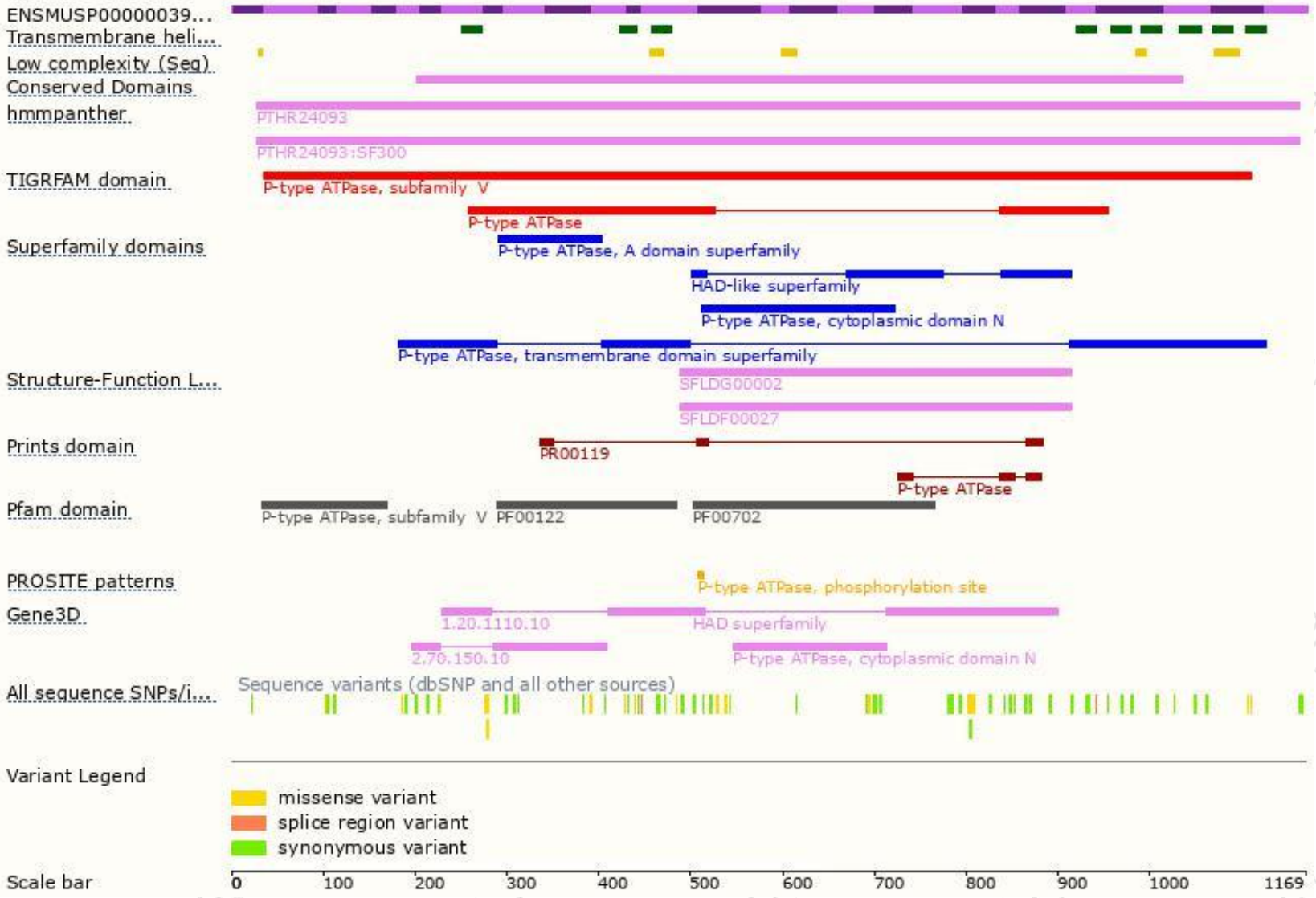
The strategy is based on the design of *Atp13a2-201* transcript,The transcription is shown below



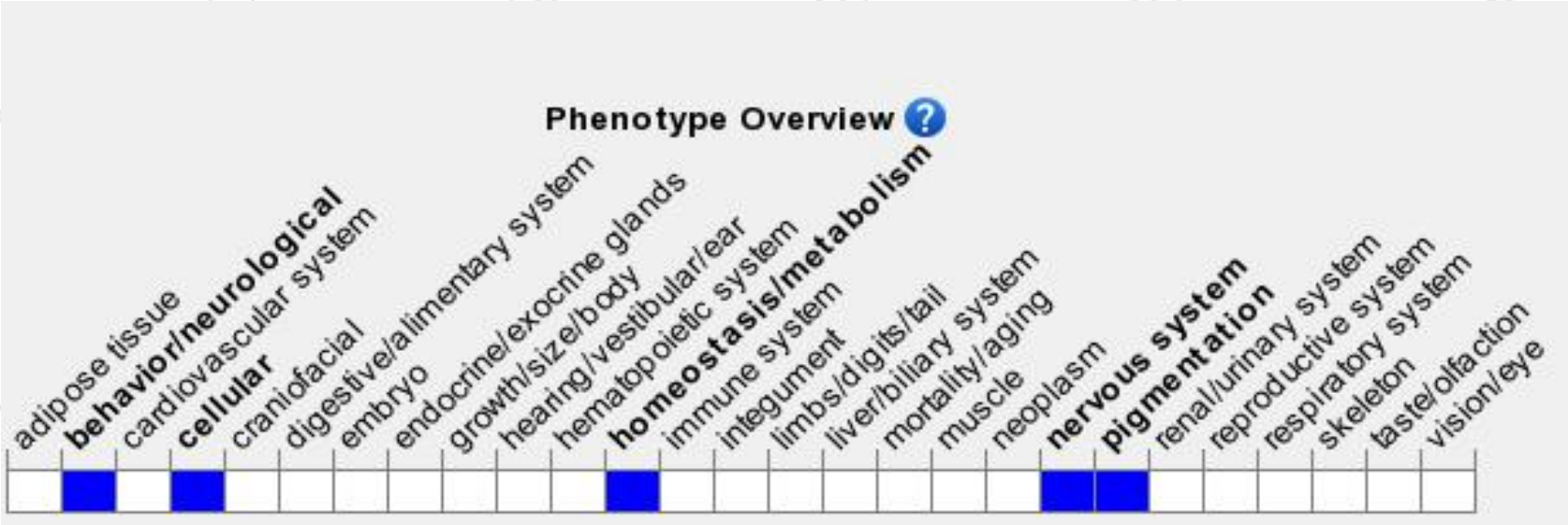
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data,Mice homozygous for a knock-out allele exhibit neuronal ceroid lipofuscinosis, synuclein accumulation and age-dependent sensorimotor deficits.

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If you have any questions, you are welcome to inquire.

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